ZYMO RESEARCH

The Beauty of Science is to Make Things Simple

INSTRUCTION MANUAL

Quick-DNA/RNA™ MagBead

Catalog Nos. **R2130, R2131**

Highlights

- High throughput (96-well) magnetic-bead based isolation of DNA and total RNA (including small/micro RNAs) from any sample source including cells, solid tissue, whole blood, biological liquids, FFPE tissue, environmental (plant/seed), swabs (stool, soil, microbial samples), *etc.*
- Perform Total Nucleic Acid Co-Purification (DNA/RNA in one elution) or DNA & RNA Parallel Purification (DNA & RNA in separate elutions from the same sample).
- High quality DNA & RNA is ready for use in any downstream application. *DNase I included.*

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ZYMO RESEARCH CORP.

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Satisfaction of all Zymo Research products is guaranteed. If you are dissatisfied with this product please contact us.

Integrity of kit components is guaranteed for up to one year from date of purchase. Reagents are routinely tested on a lot-to-lot basis to ensure they provide the highest performance and reliability.

For assistance, contact us at tech@zymoresearch.com.

Plates and other consumables sold separately: 96-well **Collection Plate** (C2002; capacity is up to 1.2 ml/well), **96-Well Block** (P1001; capacity is up to 2 ml/well), **Elution Plate** (C2003), **Cover Foil** (C2007).

[1] For FFPE tissue, the **Deparaffinization Solution** (D3067-1-20) and **2X Digestion Buffer** (D3050-1-20) are sold separately.

[1] For plant samples, the **ZR-96 BashingBead™ Lysis Rack (2.0 mm**) (S6002-96-2) and **ZR BashingBead™ Lysis Tubes (2.0 mm)** (S6003-50) are sold separately.

[1] For microbial samples, the **ZR-96 BashingBead™ Lysis Rack (0.5 mm**) (S6002-96-1) and **ZR BashingBead™ Lysis Tubes (0.1 & 0.5 mm)** (S6012-50) are sold separately.

Product Contents

***Quick*-DNA/RNA™ MagBead** (Kit Size)	**R2130** (96 Preps.)	**R2131** (4x 96 Preps.)
MagBinding Beads	3 ml	12 ml
DNA/RNA Shield™ (2X concentrate)	25 ml	125 ml
DNA/RNA Lysis Buffer	50 ml	200 ml
DNA/RNA Prep Buffer	50 ml	200 ml
MagBead DNA/RNA Wash 1 (concentrate) [1]	30 ml	120 ml
MagBead DNA/RNA Wash 2 (concentrate) [2]	20 ml	80 ml
DNase/RNase-Free Water	10 ml	2x 30 ml
DNase I[3] (lyophilized)	2	8
DNA Digestion Buffer	4 ml	4 ml
Proteinase K[4] & Storage Buffer	20 mg	4x 20 mg
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Storage Temperature - Store all kit components (*i.e.,* buffers, columns) at room temperature.

[1] Before starting, add 20 mL (R2130) or 80 ml (R2131) of isopropanol to the **MagBead DNA/RNA Wash 1** concentrate.
[2] Before starting, add 30 mL (R2130) or 120 ml (R2131) of isopropanol to the **MagBead DNA/RNA Wash 2** concentrate.
[3] Prior to use, reconstitute the lyophilized **DNase** I with 275 µl **DNase/RNase-Free Water**. Mix by gentle inversion. Store aliquots at -20ºC.
[4] Prior to use, reconstitute the lyophilized **Proteinase K** with 1040 µl **Proteinase K Storage Buffer.** Vortex to dissolve. Store at -20ºC.

Specifications

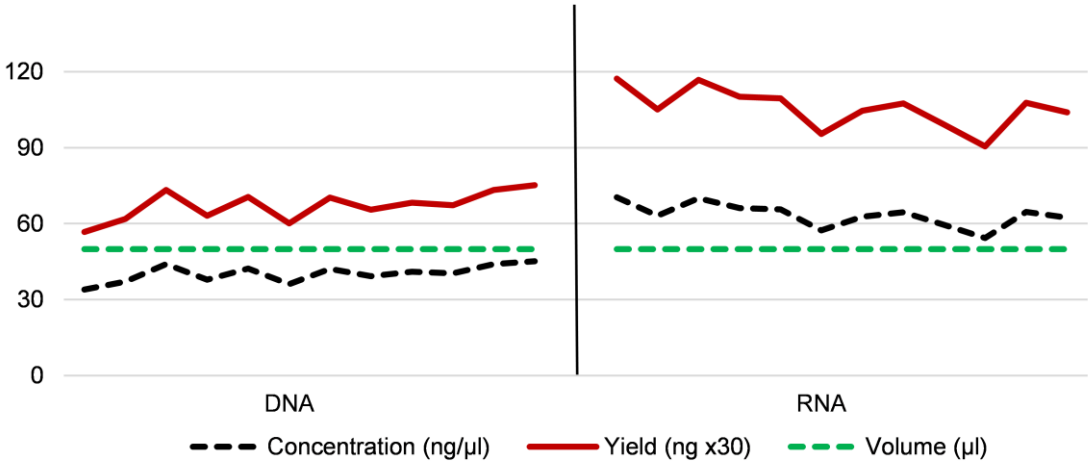
- **Sample Sources** – Any cells, solid tissue, whole blood, biological fluids, FFPE tissue, environmental (plant/seed), swabs (stool, soil, microbial samples), samples stored in DNA/RNA Shield™, *etc.*[1]
- **Sample Preservation** – **DNA/RNA Shield™** lyses cells, inactivates nucleases and infectious agents and is ideal for safe sample storage and transport at ambient temperatures (page 6).
- **Size Limits** – Capable of recovering genomic DNA up to and above 40 kb. In most instances, mitochondrial and viral DNA (if present) will also be recovered. Total RNA ≥17 nucleotides can be recovered.
- **Purity** – High quality DNA and RNA (A_{260}/A_{280} >1.8, A_{260}/A_{230} >1.8) are recovered.
- **Recovery** – The DNA/RNA binding capacity is ~20 µg (per 30 µl **MagBinding Beads**).
- **Storage** – DNA and RNA eluted with **DNase/RNase-Free Water** (provided) can be stored at ≤-70ºC. The addition of RNase inhibitors is highly recommended for prolonged storage.
- **Equipment Needed** – Centrifuge fitted with a 96 well microplate carrier, 96 well magnetic stand, 96 well plate heat block, liquid handler or other robotic sample processer.

Product Description

The ***Quick*-DNA/RNA™ MagBead** kit provides a high-throughput, magnetic bead-based purification of both high-quality DNA and total RNA (including small RNAs) from the same starting sample. The provided **DNA/RNA Shield™** inactivates infectious agent and is ideal for sample storage at ambient temperatures. The extraction method has the option to recover total nucleic acids in one elution or DNA & RNA in separate elutions without the use of phenol. It is eluted into ≥50 µl of DNase/RNase-free water and is ready for any downstream application including RT-PCR, sequencing, *etc.*

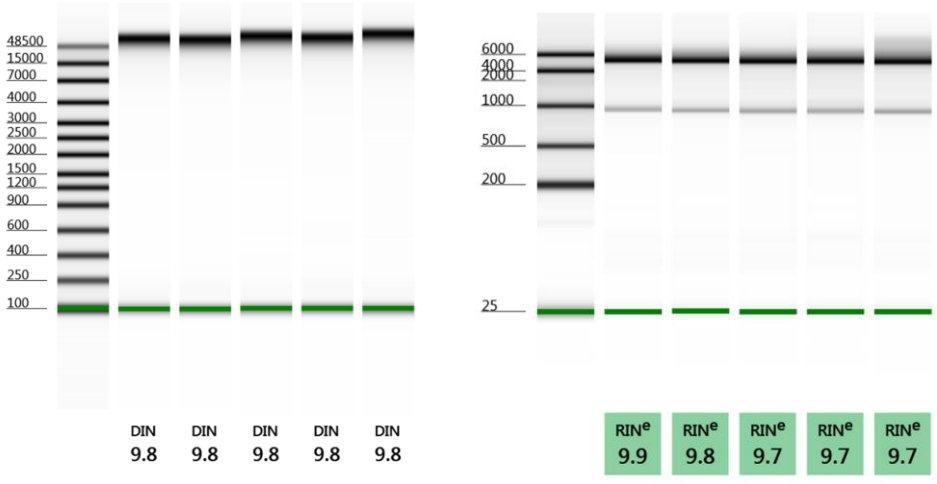
For **Assistance**, please contact Zymo Research Technical Support at 1-888-882-9682 or e-mail tech@zymoresearch.com.

Reproducible Sample Processing



Concentration, yield, and elution volume across replicate samples extracted with the ***Quick*-DNA/RNA™ MagBead** are reproducible and consistent. DNA and RNA were purified from HeLa cells (2.5 x 10^5/well).

High Quality DNA & RNA



DNA (left) and RNA (right) quality assessed using Agilent 2200 TapeStation. DNA and RNA were purified from HeLa cells using the ***Quick*-DNA/RNA™ MagBead**.

Reagent Preparation

- ✓ Add 20 ml (R2130) or 80 ml (R2131) isopropanol to the **MagBead DNA/RNA Wash 1** concentrate.
- ✓ Add 30 ml (R2130) or 120 ml (R2131) isopropanol to the **MagBead DNA/RNA Wash 2** concentrate.
- ✓ Add 1040 µl **Proteinase K Storage Buffer** per vial to reconstitute the lyophilized **Proteinase K** at 20 mg/ml. Vortex to dissolve. Store at -20ºC.
- ✓ Prepare **DNase I Reaction Mix** (according to the example below; scaled as needed).

DNase I (250 U/vial; lyophilized)	DNase/RNase-Free Water	DNA Digestion Buffer
1 vial	2.3 ml	0.3 ml
2 vials (96-well plate)	4.6 ml	0.6 ml

1. Reconstitute **DNase I** with **DNase/RNase-Free Water** (table above), transfer into an RNase-free tube (*e.g.*, 15 ml conical tube; not provided) and mix by inversion.

   At this point, aliquots can be stored frozen at -20ºC.
2. Add **DNA Digestion Buffer** (table above) and mix by inversion, then place on ice until ready to use. Add 50 µl **DNase I Reaction Mix** per sample during RNA Purification.

The lyophilized **Proteinase K** and **DNase I** are stable as shipped.

Unit definition – one unit increases the absorbance of a high molecular weight DNA solution at a rate of 0.001 A260 units/min/ml of reaction mixture at 25°C.

Protocol

The isolation consists of two steps: (I) Sample Preparation & (II) DNA/RNA Purification

(I) Sample Preparation

All centrifugation steps should be performed at 10,000 - 16,000 *x g* for 30 seconds unless specified.
All steps should be performed at room temperature (20-30ºC) unless specified.

Cells

1. Pellet up to 10^6 mammalian cells (≤500 *x g* for 1 minute), remove the supernatant and resuspend the cell pellet in 200 µl **DNA/RNA Shield**™ (1X)[1]. Proceed to Total Nucleic Acid (Page 5) or DNA & RNA Purification (page 6).

Solid Tissue & Blood Cells (PBMCs, WBCs)

1. Mechanically homogenize ≤5 mg solid tissue and add 200 µl **DNA/RNA Shield**™ (1X)[1]. For blood cells, buffy coat and pelleted PAXgene™ samples from ≤1 ml blood, resuspend in 200 µl **DNA/RNA Shield**™ (1X).
2. Add 10 µl **Proteinase K**. Mix and incubate at room temperature (20-30ºC) for 30 minutes.
3. Proceed to Total Nucleic Acid (Page 5) or DNA & RNA Purification (page 6).

Urine[2]

1. Generate pellet from up to 40 ml urine by adding 70 µl **Urine Conditioning Buffer** for every 1 ml of urine and mix by vortexing. Centrifuge at 3,000 *x g* for 15 minutes. Discard the supernatant and leave up to 50 µl pellet.
2. Add 150 µl **DNA/RNA Shield**™ (1X)[1] and resuspend the pellet by pipetting.
3. Add 10 µl **Proteinase K**. Mix and incubate at room temperature (20-30ºC) for 30 minutes.
4. Proceed to Total Nucleic Acid (Page 5) or DNA & RNA Purification (page 6).

Notes:

[1] To prepare 1X solution, mix equal amounts of the supplied 2X concentrate with nuclease-free water (not provided).

[2] Warm up urine sample at 37 ˚C for 5-10 minutes if the urine is visually cloudy (salt precipitation). Samples containing bacterial contamination will not clear.

After adding **Urine Conditioning Buffer** (sold separately; D3061-1-140), urine can be stored for up to 1 month at ambient temperature. Prior to processing, mix the sample thoroughly by vortexing.

(I) Sample Preparation (continued)n

Whole Blood (Mammalian)

1. Add 200 µl **DNA/RNA Shield™** (2X concentrate) to each 200 µl sample and mix thoroughly.
2. For every 400 µl of reagent/sample mixture, add 10 µl **Proteinase K** and mix thoroughly. Incubate at room temperature (20-30ºC) for 30 minutes.
3. Add 1 volume of isopropanol and mix well.
4. Proceed to Total Nucleic Acid (Page 5, Step 3). DNA & RNA Purification is not compatible.

FFPE Tissue

1. Remove (trim) excess paraffin wax from ≤5 mg FFPE tissue and transfer into a new tube.
2. Add 400 µl **Deparaffinization Solution**[1] to the sample. Incubate at 55ºC for 1 minute. Vortex briefly. Remove the **Deparaffinization Solution**.
3. Add 95 µl **DNase/RNase-Free Water**, 95 µl **2X Digestion Buffer**, and 10 µl **Proteinase K**.
4. Incubate at 55ºC for 1 hour. Incubate at 94ºC for 20 minutes to de-crosslink the sample.
5. Centrifuge to remove insoluble debris and transfer supernatant to a new tube. Proceed to Total Nucleic Acid (Page 5) or DNA & RNA Purification (page 6).

Environmental (Plant/Seed, *etc.*)

1. Add up to 50 mg plant material and 750 µl **DNA/RNA Shield™** (1X)[3] to a bead beating tube[2].
2. Secure in a bead beater fitted with the appropriate holder assembly for your bead beating module and process at maximum speed for 1 minute.[4]
3. Add 10 µl **Proteinase K**. Mix and incubate at room temperature (20-30ºC) for 30 minutes.
4. Centrifuge to pellet debris. Transfer 200 µl supernatant to a new tube. Proceed to Total Nucleic Acid (Page 5) or DNA & RNA Purification (page 6).

Swabs (Stool, Soil, Microbial samples, *etc.*)

1. Add 750 µl **DNA/RNA Shield™** (1X)[3] to a swab sample (up to 50 mg stool or soil) and mix by vortexing. Centrifuge to pellet debris. Transfer 200 µl supernatant to a new tube.

   Optional: To achieve unbiased lysis of different organisms, including hard-to-lyse microbes, use bead beating tubes[5]. Secure in a bead beater fitted with the appropriate holder assembly for your bead beating module and process at maximum speed for 5 minutes.[4]
2. Add 10 µl **Proteinase K**. Mix and incubate at room temperature (20-30ºC) for 30 minutes.
3. Proceed to Total Nucleic Acid (Page 5) or DNA & RNA Purification (page 6).

Notes:

Compatible with commonly used anticoagulants (*i.e.,* EDTA, citrate, heparin).

[1] **Deparaffinization Solution** (D3067-1-20) and **2X Digestion Buffer** (D3050-1-20) are sold separately.

[2] **ZR-96 BashingBead™ Lysis Rack (2.0 mm**) (S6002-96-2) and **ZR BashingBead™ Lysis Tubes (2.0 mm)** (S6003-50) are sold separately.

[3] To prepare 1X solution, mix equal amounts of the supplied 2X concentrate with nuclease-free water (not provided).

[4] Processing time will vary based on sample input and bead beater. Times may be as little as 5 minutes using high-speed cell disrupters (*e.g.* FastPrep®-96) or as long as 20 minutes using lower speeds.

[5] **ZR-96 BashingBead™ Lysis Rack (0.1 & 0.5 mm**) (S6002-96-3) and **ZR BashingBead™ Lysis Tubes (0.1 & 0.5 mm)** (S6012-50) are sold separately.

Notes:

[1] For all buffer additions and incubation steps, **mix well** by pipetting up and down several times and/or (if available) by vortexing at ~1,000 rpm.

[2] This protocol can be performed using a 96-well **Collection Plate** (C2002; capacity is up to 1.2 ml/well), **96-Well Block** (P1001; capacity is up to 2 ml/well) or a nuclease-free tube (not provided).

[3] **MagBinding Beads** settle quickly, ensure that beads are kept in suspension while dispensing.

[4] Magnetic stand (manual processing) or strong-field 96-well magnetic stand (*i.e.,* **ZR-96 MagStand**, P1005).

[5] Some beads will adhere to the sides of the well. When removing the supernatant, aspirate slowly to allow these beads to be pulled to the magnet as the liquid level is lowered.

[6] Beads will change in appearance from glossy black when still wet to a dull brown when fully dry.

(II) Total Nucleic Acid (DNA/RNA)

1. Add 200 µl (1 volume) **DNA/RNA Lysis Buffer**[1] to the sample and mix well[2].
2. Add 400 µl ethanol (95-100%) to the sample and mix well.
3. Add 30 µl **MagBinding Beads**[3] and mix well for 20 minutes.
4. Transfer the plate (tube) to the magnetic stand[4] (sold separately) until beads have pelleted, then aspirate[5] and discard the cleared supernatant.
5. Add 500 µl **MagBead DNA/RNA Wash 1** and mix well.
6. Transfer the plate (tube) to the magnetic stand until beads have pelleted, then aspirate and discard the cleared supernatant.
7. Add 500 µl **MagBead DNA/RNA Wash 2** and mix well.
8. Transfer the plate (tube) to the magnetic stand until beads have pelleted, then aspirate and discard the cleared supernatant.
9. Add 500 µl ethanol (95-100%) and mix well.
10. Transfer the plate (tube) to the magnetic stand until beads have pelleted, then aspirate and discard the cleared supernatant.
11. Repeat the wash (steps 9-10).

    To remove DNA and extract RNA alone, perform DNase treatment in RNA Purification (page 6, steps 11-13).
12. To dry the beads, transfer the plate (tube) to a heated element and incubate at 55ºC for 30 minutes or until dry.[6]
13. To elute DNA/RNA from the beads, add 50 µl **DNase/RNase-Free Water** and mix well for 5 minutes.
14. Transfer the plate (tube) to the magnetic stand until beads have pelleted, then aspirate and dispense the eluted DNA/RNA to a new plate (tube).

    The eluted total nucleic acid can be used immediately or stored at ≤-70ºC.

    Use the **Cover Foil** to prevent evaporation.

(II) DNA & RNA Parallel Purification

1. Add 500 µl (2.5 volumes) **DNA/RNA Lysis Buffer**[1] to the 200 µl sample and mix well[2].
2. Add 30 µl **MagBinding Beads**[3] and mix well for 20 minutes.
3. Transfer the plate (or tube) to the magnetic stand[4] (sold separately) until beads (DNA) have pelleted, then transfer[5] the cleared supernatant (RNA) into a new plate (tube).

DNA Purification (Beads)

4. Add 500 µl **MagBead DNA/RNA Wash 1** and mix.
5. Transfer the plate (tube) to the magnetic stand until beads have pelleted, then aspirate and discard the cleared supernatant.
6. Add 500 µl **MagBead DNA/RNA Wash 2** and mix. Pellet the beads and discard the supernatant.
7. Add 500 µl ethanol (95-100%) and mix. Pellet the beads and discard the supernatant.
8. Repeat wash step 7.
9. Transfer the plate (tube) to a heated element and incubate at 55ºC for 30 minutes or until dry.[6]
10. Add 50 µl **DNase/RNase-Free Water** and incubate for 5 minutes while mixing.
11. Transfer the plate (tube) to the magnetic stand until beads have pelleted, then aspirate and dispense the eluted DNA to a new plate (tube).

RNA Purification (Supernatant)

4. Add 700 µl (1 volume) ethanol (95-100%) to the supernatant and mix.
5. Add 30 µl/well **MagBinding Beads** and mix well for 10 minutes.
6. Transfer the plate (tube) to the magnetic stand until beads have pelleted, then aspirate and discard the cleared supernatant.
7. Add 500 µl **MagBead DNA/RNA Wash 1** and mix. Pellet the beads and discard the supernatant.
8. Add 500 µl **MagBead DNA/RNA Wash 2** and mix. Pellet the beads and discard the supernatant.
9. Add 500 µl ethanol (95-100%) and mix. Pellet the beads and discard the supernatant.
10. Repeat wash step 9.
11. Add 50 µl **DNase I Reaction Mix** and mix gently. Incubate at room temperature for 10 minutes.
12. Add 500 µl **DNA/RNA Prep Buffer** and mix for 10 minutes. Pellet the beads and discard the supernatant.
13. Repeat the wash (steps 9-10).
14. Transfer the plate (tube) to a heated element and incubate at 55ºC for 30 minutes or until dry.[6]
15. Add 50 µl **DNase/RNase-Free Water** and incubate for 5 minutes while mixing.
16. Transfer the plate (tube) to the magnetic stand until beads have pelleted, then aspirate and dispense the eluted RNA to a new plate (tube).

The eluted DNA & RNA can be used immediately or stored at ≤-70ºC.

Use the **Cover Foil** to prevent evaporation.

Notes:

[1] For all buffer additions and incubation steps, **mix well** by pipetting up and down several times and/or (if available) by vortexing at ~1,000 rpm.

[2] This protocol can be performed using a 96-well **Collection Plate** (C2002; capacity is up to 1.2 ml/well), **96-Well Block** (P1001; capacity is up to 2 ml/well) or a nuclease-free tube (not provided).

[3] **MagBinding Beads** settle quickly, ensure that beads are kept in suspension while dispensing.

[4] Magnetic stand (manual processing) or strong-field 96-well magnetic stand (*i.e.,* **ZR-96 MagStand**, P1005).

[5] Some beads will adhere to the sides of the well. When removing the supernatant, aspirate slowly to allow these beads to be pulled to the magnet as the liquid level is lowered.

[6] Beads will change in appearance from glossy black when still wet to a dull brown when fully dry.

Appendices

Compatibility with DNA/RNA Shield™ Collection Devices

Blood	Swabs	Stool	Tissue
Fresh EDTA Citrate Heparin	Mouth Nose Throat Fluid	Virus Microbe Host	Animal Plant Insect Microbe
			
16x100 mm vacuum tube 3 ml draw #R1150 (50 pack)	**12x80 mm tube + swab** w/ 1 ml reagent #R1106 (10 pack) #R1107 (50 pack) w/ 2 ml reagent #R1108 (10 pack) #R1109 (50 pack)	**20x76 mm scoop tube** w/ 9 ml reagent #R1101 (10 pack)	**2 ml lysis tube** 1 ml reagent #R1102 (50 pack) Microbe (+beads) #R1103 (50 pack) Microbe w/ swab #R1104 (50 pack) Tissue #R1105 (50 pack)

Sample Preservation in DNA/RNA Shield™

DNA/RNA Shield™ effectively lyses cells, inactivates nucleases and infectious agents and is ideal for sample storage/transport at ambient temperatures prior to nucleic acid purification.

Liquid samples: Mix an equal volume DNA/RNA Shield™ (2X concentrate) and sample.
Solid samples: Submerge sample (not to exceed 10% (v/v or w/v) in DNA/RNA Shield (1X).

Mix well/homogenize sample prior to storage.

Samples in DNA/RNA Shield™ can be stored at ambient temperature (4-30ºC) for a month, 3 days at 37ºC or long term (>1 year) at -20ºC or below.

Automation Scripts

The ***Quick*-DNA/RNA™ MagBead** is compatible with automated platforms. For automation scripts and related technical support, email tech@zymoresearch.com. In the subject line, please include "Automation Scripts", instrument used and the product catalog number.

Ordering Information

Product Description	Catalog No.	Kit Size
***Quick*-DNA/RNA™ MagBead**	R2130 R2131	96 Preps. 4x 96 Preps.

For Individual Sale	Catalog No.	Amount
MagBinding Beads	D4100-2-3 D4100-2-12 D4100-2-24	3 ml 12 ml 24 ml
DNA/RNA Shield™ (2X concentrate)	R1200-25 R1200-125	25 ml 125 ml
DNA/RNA Lysis Buffer	D7001-1-50 D7001-1-200	50 ml 200 ml
DNA/RNA Prep Buffer	D7010-2-50 D7010-2-200	50 ml 200 ml
MagBead DNA/RNA Wash 1 (concentrate)	R2130-1-30 R2130-1-120	30 ml 120 ml
MagBead DNA/RNA Wash 2 (concentrate)	R2130-2-20 R2130-2-80	20 ml 80 ml
DNase/RNase-Free Water	W1001-1 W1001-4 W1001-6 W1001-10 W1001-30	1 ml 4 ml 6 ml 10 ml 30 ml
DNase I Set (lyophilized) DNase I (250 U) & DNA Digestion Buffer (4 ml)	E1010	1 set
Proteinase K (lyophilized) (supplied with Proteinase K Storage Buffer)	D3001-2-5 D3001-2-20	5 mg set 20 mg set
Collection Plate	C2002	2 plates
Elution Plate	C2003	2 plates
96-Well Plate Cover Foil	C2007-2 C2007-4 C2007-8	2 4 8
ZR-96 MagStand	P1005	1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